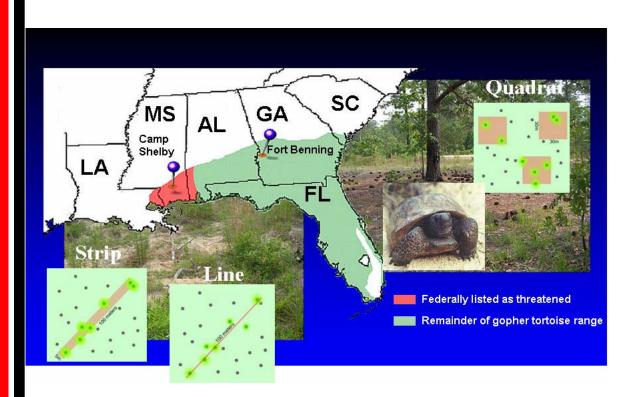


Analysis of Gopher Tortoise Population Estimation Techniques

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ABSTRACT: Estimating threatened and endangered species (TES) population trends is essential to be able to track a species' recovery progress. Natural resources managers on military installations use a wide variety of survey and monitoring methods, with incomplete knowledge toward their accuracy. Using the gopher tortoise (*Gopherus polyphemus*) as the test case species, this report examines TES population estimation techniques for accuracy. Managers are cautioned to not accept surveys with unchallenged assumptions of total observability of burrows, and to not convert survey results to tortoise numbers utilizing published correction factors that may not be applicable to a local survey site.

The versatility of the quadrat (plot) sampling method recommends it as a default technique that can be used in many sampling circumstances with reasonable expectations of accuracy. Strip transects with reasonable estimates of detectability can offer similar utility when habitat conditions make them appropriate.

Previous studies comparing burrow survey methodologies have not adequately addressed the issue of detectability, which presents a weakness in current population estimation and total count techniques. It is therefore recommended to undertake a rigorous field test of these techniques to remove the detectability weakness.

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Preface

This study was conducted for the Department of the Army, Office of the Director of Environmental Programs under project A896, "Base Facilities Environmental Quality." The technical monitor was Bill Woodson, DAIM-ED-N.

The work was performed by the Ecological Processes Branch (CN-N) of the Installations Division (CN), Construction Engineering Research Laboratory (CERL). The CERL Principal Investigator was William D. Meyer. Part of this work was done by Dr. Raymond R. Carthy, Dr. Madan K. Oli, and Dr. John B. Wooding, U.S. Geological Survey, Florida Cooperative Fish & Wildlife Research Unit at the University of Florida and Dr. Joan E. Berish, under MIPR W81EWF32592004. The technical editor was Gloria J. Wienke, Information Technology Laboratory. Alan B. Anderson is Chief, CEERD-CN-N, and L. Michael Golish is Acting Chief, CEERD-CN. The associated Technical Director was William D. Severinghaus, CEERD-CV-T. The Acting Director of CERL is Dr. Ilker Adiguzel.

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1 Introduction

Background

The U.S. Army Engineer Research and Development Center's Construction Engineering Research Laboratory (ERDC/CERL) in Champaign, IL, is engaged in research to examine threatened and endangered species population estimation techniques for accuracy in a variety of vegetative site conditions. Estimating threatened and endangered species (TES) population trends is essential to be able to track a species' recovery progress. Yet, for many TES this continues to be a difficult problem due to the wide variety of survey and monitoring methods employed for this purpose with incomplete knowledge toward their accuracy. This is especially true on military installations. Human and equipment resources vary according to the installation, as do available funds for studies. In order for installations to be able to make an informed choice about which technique to best apply, they need information regarding the accuracy of population estimation techniques given differing levels of vegetative site condition.

To develop this information the gopher tortoise (Gopherus polyphemus) was selected as the test case species. The gopher tortoise was selected because it is listed as threatened in the portion of its range west of the Mobile and Tombigbee rivers in Alabama, Mississippi, and Louisiana, and considered a species at risk in the eastern portion of its range. In a proactive effort to try and keep the species from becoming listed in the eastern portion of its range, the Army wants to be prepared with the best scientific methods and techniques available to meet this challenge. The gopher tortoise is a terrestrial reptile that was once found throughout the southeastern United States from North Carolina into Texas. However, due to numerous factors including human and animal predation and habitat loss, they have been in decline for the past several decades. Populations often occur on military bases, where disturbance of the species is minimized through management of the species. Tortoises prefer open-canopied habitats with ample herbaceous ground vegetation for forage. Clearing trees to make openings at tank firing points and removing understory to facilitate maneuver training has created good habitat, and the tortoises have moved in. Their presence is a potential conflict with the training mission and makes them an important mitigation priority. ERDC/CERL in cooperation with the U.S. Geological Survey, Florida Fish and Wildlife Cooperative Extension Unit at the University of Florida performed research comparing the strengths and weaknesses of go-

pher tortoise survey and monitoring techniques. The results of this study identify the strengths and weaknesses of each technique across varying site conditions with suggestions for new protocols to reduce bias and incorporate detection statistics. Lessons learned from this research will serve in the design of similar studies on surveying and monitoring for other high priority TES on military lands.

Objective

The objective of this research is to analyze the accuracy of species population estimation techniques. For this study **Accuracy** is defined as the highest degree of validity that can be achieved in the result. Using the gopher tortoise as a test case, this research will prepare a detailed analysis of current population estimation methods with the intent of identifying the accuracy of each technique. This research will help natural resource managers on Department of Defense (DoD) and non-DoD lands to select and use the most effective inventory and monitoring techniques.

Approach

This research will prepare a detailed analysis of all current population estimation methods with the intent of identifying the accuracy of each technique. From these techniques the researchers will be asked to recommend up to three techniques that maximize accuracy over the widest range of site conditions. The researchers will also be asked to compare and contrast the techniques and rate them in relation to one another. The researchers conducting this study should also suggest improvements to these techniques or suggest entirely new techniques that might further improve the accuracy of population estimation techniques. To accomplish this research, the research team will conduct a thorough literature review of previous research along with telephone interviews of principals and practioners who regularly conduct gopher tortoise population surveys. The research team will also conduct computer simulation modeling of population estimation based on existing datasets from two military locations across the home range of the gopher tortoise. To be most effective this research must succeed in meeting the following criteria or suggest ways in which the following criteria can be met through the introduction of new methods or other refinements.

- 1. Identify the most robust survey techniques given differing levels of vegetative site condition.
- 2. Identify the level of effort (timing, quantity, and frequency) required to achieve the greatest level of data validity for each technique.

3. Identify the level of each techniques' standardized application (fieldwork, database management, and statistical analysis) that would allow identification of population trends.

- 4. Provide clear protocols and guidelines that would ensure appropriate application of these techniques in the field.
- 5. Ensure that all endorsed methods meet U.S. Fish and Wildlife Service (USFWS) Gopher tortoise monitoring requirements.

Mode of Technology Transfer

This report has informed an ongoing study to produce guidelines for gopher tortoise inventory and monitoring.

This report will be made accessible through the World Wide Web (WWW) at URL: http://www.cecer.army.mil

2 Review of Gopher Tortoise Survey Techniques

Gopher tortoises are terrestrial, burrowing turtles restricted to Florida and the Coastal Plain of South Carolina, Georgia, Alabama, Mississippi, and Louisiana (Auffenberg and Franz 1982). Historically, the species was relatively common and widespread in fire-maintained pine savannas and coastal scrub communities, but they are now uncommon due mostly to agricultural, silvicultural, and urban land development, woodland fire suppression, and human predation for their meat (Auffenberg and Franz 1982, Lohoefener and Lohmeier 1984, U.S. Fish and Wildlife Service 1990, Berish 2001, Hermann et al. 2002). Gopher tortoises are federally listed as threatened in Louisiana, Mississippi, and western Alabama (Federal Register 7 July 1987). As of February 2004, their state protected status varied by state in the areas east of the Tombigbee and Mobile rivers, which is the line in Alabama separating the eastern portion of their range from the western portion. In South Carolina they are endangered. In Georgia they are threatened. In Florida they are a species of special concern. In eastern Alabama they are protected.

Biology Relevant to Estimates of Abundance

Gopher tortoises are reclusive animals, spending most of their lives sheltered underground in their burrows (Smith 1992, Wilson et al. 1994, Eubanks et al. 2003). The burrow entrances of adults are conspicuous because of the large, flattened mound of excavated, bare soil (commonly referred to as the apron) and the halfmoon shaped entrance. Tortoises are normally solitary dwellers, but double occupancy of burrows has been observed (Douglass 1986, Diemer 1992a). Burrow depth averages 4.5 m (Hansen 1963). Burrow entrances are about twice as wide as high (Hallinan 1923, Hansen 1963, Doonan and Stout 1994), and the dimensions closely match the dimensions of the occupant (Hansen 1963, Alford 1980, Wilson et al. 1991, Doonan and Stout 1994). The latter is a characteristic used by Alford (1980) to study tortoise demographics without observing the tortoises themselves. Gopher tortoises show a preference for burrowing in well-drained, sandy soils on higher ground (Garner and Landers 1981, Auffenberg and Franz 1982), presumably because of easier digging and less chance of burrow flooding. However, they are capable of burrowing in denser, clay soils (Guyer and Hermann 1997, Wester 2003), and they can tolerate burrow flooding by surfacing to breathe (Means 1982).

Once excavated, an adult burrow may remain in use for 3 to 12 years (Guyer and Hermann 1997); burrows are used for longer periods under stable resource conditions, and for shorter periods when resources and the dependent tortoises are transitory (Guyer and Hermann 1997, Aresco and Guyer 1999). Unused burrows eventually fill in with soil and seed over, but they can be recognized as tortoise burrows for months, and in some cases for years, after they have been abandoned. Abandoned burrows can be reopened and used again by tortoises, and armadillo use of tortoise burrows (Guyer and Hermann 1997) may cause a burrow to be considered "active."

Tortoises prefer habitat with direct sun at ground level (Auffenberg and Franz 1982, Stewart et al. 1993, McCoy and Mushinsky 1995, Aresco and Guyer 1999, Boglioli et al. 2000, Waddle 2000). Solar energy is used for thermoregulation and egg incubation. Also, tortoises are grazers (Garner and Landers 1981, Lohoefener and Lohmeier 1981, Macdonald and Mushinsky 1988), and food abundance at ground level is greater in sunny habitats. The quantity of sun at ground level and the ensuing plant communities are probably more important to tortoises than soil types, provided the soil is at least minimally suitable for burrowing. Tortoises tend to occur in aggregations referred to as colonies (Auffenberg and Franz 1982). Alford (1980) defined a colony as ≥5 burrows/ha. In the western portion of their range, a colony is defined for regulatory purposes as 2 or more active or inactive burrows within 600 feet of each other (USFWS). Colonies form due to conspecific attraction, but concentrations may also form because of habitat limitations, with tortoises concentrating their burrows in patches of sunlight such as those found in forest openings and along power line rights-of-way and roads.

Gopher tortoises are diurnal (Hallinan 1923, Douglass and Layne 1978) and their activity varies seasonally, with the lowest activity occurring in winter and the greatest in summer (Douglass and Layne 1978, McRae et al. 1981, Diemer 1992a, 1992b). In the winter they may only emerge to bask at the burrow mouth on the warmest days (Douglass and Layne 1978). In summer, they may emerge daily to graze (Breininger et al.1991), although some individuals may go weeks without emerging in summer.

Most summer movements are restricted to within a few dozen meters of the burrow (McRae et al. 1981, Smith 1992), and the occasional long distance movement is generally less than 500 meters (Diemer 1992a). Each tortoise uses several burrows in a year (McRae et al. 1981, Diemer 1992a, Eubanks et al. 2003), and through the use of multiple burrows, they are able to move about their home range while remaining near a burrow in case danger approaches. Male home ranges are 2 to 3 times the size of adult female ranges (Diemer 1992a, Smith et al. 1997, Eubanks et al. 2003), but neither sex could be considered wide-ranging, with adult male home ranges av-

eraging 0.9 to 1.9 ha (Diemer 1992a, Smith et al. 1997, Eubanks et al.2003) and adult female home ranges averaging 0.3 to 0.6 ha (Diemer 1992a, Smith 1992, Smith et al. 1997, Eubanks et al. 2003). There is some suggestion that the number of burrows used per tortoise declines with increasing habitat quality, perhaps because tortoises do not have to travel as far to find food and mates in higher quality habitat, and therefore they do not need as many burrows as they do in poor quality habitat where resources are more dispersed (Moler and Berish 2001).

Nesting occurs in mid-May through June (Landers et al. 1980, Smith 1992, Diemer and Moore 1994, Butler and Hull 1996). Nests may be located in burrow mounds, or away from the mound if open ground exists nearby (Hallinan 1923, Landers et al. 1980, Smith 1992, Butler and Hull 1996). Gopher tortoise mounds located near wetlands have been used by aquatic turtle species as nest sites (Landers et al. 1980). Smith (1992) observed that tortoises prefer to nest away from the burrow mound if given a choice, a behavior that may minimize the chance that a nest will be found and destroyed by predators familiar with mound-located nests. On a Georgia study area (Landers et al. 1980), 89 percent of the tortoise nests were destroyed by predators. Eggs in the nests that escape predation hatch after an incubation period of approximately 3 to 3.5 months (Landers et al. 1980, Smith 1992, Butler and Hull 1996). Tortoises are capable of digging their first burrows as hatchlings (Landers et al. 1980, Doonan and Stout 1994, Butler and Sowell 1996), but they may also shelter in adult burrows or under litter their first year (Douglass 1978). Hatchling burrow entrances are approximately 5 cm wide (Doonan and Stout 1994) with inconspicuous quantities of excavated dirt. This contrasts to the entrances of adult burrows, which are at least 23 cm wide (U.S. Fish and Wildlife Service 1990), with mounds of excavated soil usually greater than 1 square meter.

Examples of Tortoise Censusing Projects

People have expended considerable effort trying to estimate gopher tortoise abundance for three reasons: (1) status assessments (e.g., Logan 1981, Auffenberg and Franz 1982, Lohoefener and Lohmeier1984, Spillers and Speake 1988, McCoy and Mushinsky 1991, 1992a, Hermann et al. 2002); (2) research projects (e.g., Tuberville and Dorcas 2001, Rostal and Jones 2002); and (3) compliance with endangered species laws (e.g., Wester and Swing 1992, Mann 1993, Estes and Mann 1996, Wester 2003). Surveys have focused on preferred soil types (Spillers and Speake 1988, Wester 2003), and preferred vegetative types (Auffenberg and Franz 1982, McCoy and Mushinsky 1991), but there have also been extensive surveys in which people have searched for gopher tortoises on all upland soils and all upland vegetative communities, with only water bodies and jurisdictional wetlands excluded from con-

sideration (Will McDearman, Gopher Tortoise Species Coordinator, U.S. Fish and Wildlife Service, personal communication, 2 March 2004).

Census Techniques – Overview

The various methods used to estimate gopher tortoise population size have been previously reviewed (Cox et al. 1987, Burke and Cox 1988, Burke 1990, Epperson and Heise 2001, and Moler and Berish 2001, Smith et al. 2005), and the methods can generally be reduced to the following: first, inventory and classify the burrows; and second, estimate the proportion of burrows that are occupied. This process is necessary since there are more burrows than tortoises; a ratio that varies by habitat quality, season, site, and year (Auffenberg and Franz 1982, Breininger et al. 1991, Diemer 1992b, McCoy and Mushinsky 1992b, Moler and Berish 2001, Eubanks et al. 2003). The techniques used to estimate abundance of other tortoise species, such as mark-recapture, line transect surveys of the tortoises themselves, and scat counts (e.g., Burroughs and Williams 2000, Anderson et al. 2001, Swann et al. 2002, Krzysik 2002) are not considered the best methods for obtaining abundance estimates of gopher tortoises due to assumption violations for mark-recapture methods, the fact that gopher tortoises are rarely above ground for direct counts using line transects, and rapid decomposition of scats in the southeast in comparison to arid regions where scats are more persistent.

Estimation of Burrow Abundance

Burrow searches, using either a sampling or total count, are normally conducted on foot during the growing season when tortoises are active. In one instance, a winter burrow survey was conducted in South Carolina (Tuberville and Dorcas 2001). In open habitat such as a mowed pasture, searches have been made from all-terrain vehicles or trucks. A burrow search in a Florida orange grove was made from a helicopter (Humphrey et al. 1985).

Biologists have used four primary methods to estimate abundance of gopher tortoise burrows — strip transect, line transect, quadrat count, and total count — all of which yield estimates of burrow density (burrows/area). Each method is discussed below:

Strip Transect

The strip transect method (Figure 1) involves sampling the area with a series of long, thin plots and extrapolating the results to the total area (Eberhardt 1978). Linear plots as employed in strip transect surveys are generally the most efficient plot shape for ecological sampling (Thompson 1992, Krebs 1999). Strip transects were used by Auffenberg and Franz (1982) in the first large-scale census of gopher tortoises, and their general methods have been duplicated by many (Alford 1980, Logan 1981, Spillers and Speake 1988, McCoy and Mushinsky 1991, 1992a). The transects used by Auffenberg and Franz (1982) were 150 m x 7 m, but a variety of transect lengths and widths have been used by other researchers. Transects of variable length were used by Lohoefener and Lohmeier (1984) and McCoy and Mushinsky (1991, 1992a).

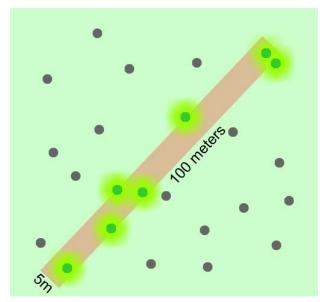


Figure 1. Strip transect survey method.

Strip transects are usually searched for burrows by one observer walking the plot centerline in one direction, with the assumption that all burrows are observed. McCoy and Mushinsky (1991) used 3 observers walking abreast in 7-m wide plots to meet this assumption. They used multiple observers because objects on the outside edge of line transects are much easier to miss, thus leading to an underestimate of density (Anderson and Pospahala 1970, Burnham and Anderson 1984). This shortcoming in the line transect method was addressed by Lohoefener and Lohmeier (1984) by varying the transect width for the conditions. They determined that the optimal plot width for the detection of \geq 98 percent of large burrows (\geq 20 cm wide) by one observer walking the centerline was either 3.2 m, 6.4 m, 9.6 m, or 12.8 m depending on vegetation density (detectability of burrows <20 cm was not measured). They chose the best width prior to beginning the surveys after inspecting the habi-

tat. They measured transect length by pacing, and plot width by perpendicular, line-of-sight angles from the observer to the burrow (Lohoefener and Lohmeier 1984).

A version of the strip transect method using randomly located transects (250 m x 20 m) as detailed by Cox et al. (1987) is currently recommended by the state of Florida for consultants evaluating development impacts on gopher tortoises (R. McCann, Biological Scientist, Florida Fish and Wildlife Conservation Commission, personal communication, 15 January 2004). This is not documented State policy, but is offered upon inquiry as a reference to an acceptable Standard Operation Procedure). They recommend the transect be searched for burrows after the centerline is measured and marked with flagging, an added step that probably increases the chances of detecting burrows because the transect is traveled twice, and from different angles.

Line Transect

The line transect method (Figure 2) using distance measurements (Burnham et al. 1980, Buckland et al. 2001) is the second commonly used approach to estimate burrow abundance. This method specifically addresses burrow detectability with the assumption that burrows are overlooked in increasing proportions away from the centerline. Using this method, an observer travels a straight line of known distance looking for burrows. The perpendicular distance or sighting angle and distance from the centerline to each burrow is measured, and through the use of welldeveloped mathematical functions, total burrow abundance is estimated. Burnham et al. (1980) recommended that abundance estimates be based on measurements to a minimum of 40 objects (i.e., burrows), and preferably 60 to 80 objects. The calculations are made with the aid of a computer and a version of the program DISTANCE* (Thomas et al. 2003), although Lohoefener (1990) developed his own program for the calculations. The line transect method with distance measurements has been used in several gopher tortoise population estimation projects (Doonan 1986, Lohoefener 1990, Mann 1993, Franz et al. 1998, Epperson 1997, Hermann et al. 2002). It was attempted by Franz et al. (1998) but was dropped because too few burrows were observed from the transect line to meet sample size requirements, due to thick vegetation and a low density tortoise population. For example, in one portion of the study area, 20,330 m of transects yielded only 6

^{*} Citing trade names does not constitute endorsement by the Department of Defense or the U.S. Army. DISTANCE is available from http://www.ruwpa.st-and.ac.uk/distance/

burrows (A. Kinlaw, PhD candidate, Department of Wildlife, University of Florida, personal communication, 4 March 2004).

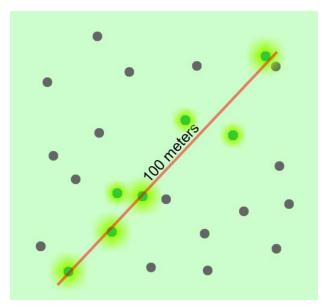


Figure 2. Line transect survey method.

Lohoefener (1990) conducted a pilot study in southern Alabama to evaluate the line transect method for gopher tortoise abundance estimation, and he provides a detailed description of his technique. The 149-ha study area was divided into 11 sampling units, and a transect with multiple segments was run through each unit. The effective transect width of 32 m was a 2.5-fold increase in width over the widest strip transect (12.8 m) that he and Lohmeier (1984) used in an earlier survey in Mississippi, thus more than doubling the area surveyed per transect length. The transects were systematically located from a randomly chosen starting point. The transects were laid out first, and then surveyed by one observer. Transect segments were spaced a minimum of 50 m apart to avoid counting burrows twice from adjoining segments. Transect length for the total area was arbitrarily set at 10,000 m before the study began, and it was exceeded by 1,200 m by study's end. Lohoefener (1990) sampled a total area of 35.8 ha with 11,200 m of transects, equaling a 24 percent sample of the study area. Transects were perpendicular to topographic contours. Distance measurements were made for the 89 burrows observed. The sample size satisfied the requirements of Burnham et al. (1980); however, sample sizes for each unit fell below the recommended size of 40 burrows, with numbers ranging from 0 to 32. The coefficient of variation (c.v.) of the abundance estimate was 26 percent for all 11 transects, satisfying Lohoefener's (1990) requirement that the c.v. be ≤33 percent. The c.v. for individual transects was not reported. He estimated that 19,000 m of transects would be needed for a 20 percent c.v. Based on the conditions of Lohoefener's (1990) pilot study, 11,200 m of transects were an adequate sample size for 149 ha.

Quadrat Count

Quadrat counts (Krebs 1999; Figure 3) using randomly located plots is the third burrow inventory method. This is a standard technique in plant surveys, but its use for inventorying gopher tortoise burrows has been restricted to two Florida surveys, both of which occurred in dense scrub habitat (Breininger et al. 1991, Franz et al. 1998). The vegetation was found to be too dense to efficiently search using strip or line transects as was intended when the surveys began (A. Kinlaw, 4 March 2004). Since extensive, time-consuming searches were judged necessary due to habitat conditions, the authors decided to employ large plots — 100 m x 100 m (Franz et al. 1998) and 50 m x 30 m (Breininger et al.1991). Even with this step, A. Kinlaw (pers. comm.) estimated that 5 percent of the burrows were overlooked in the 100-m x 100-m plots due to thick vegetation. The relationship between vegetation density and burrow detectability has been noted in other studies (Lohoefener and Lohmeier 1984, Burke and Cox 1988, Diemer 1992b). Tuberville and Dorcas (2001) conducted their surveys in winter with the assumption that dormant vegetation led to increased burrow detection. Diemer (1992b) observed that juvenile burrows were easier to see in burned rather than unburned areas. The benefits of fire at increasing burrow detection was also noted by Smith (1992), Mann (1993), and Moler and Berish (2001). In fact, Mann (1993) and Moler and Berish (2001) recommend using prescribed fire to clear vegetation prior to surveys.

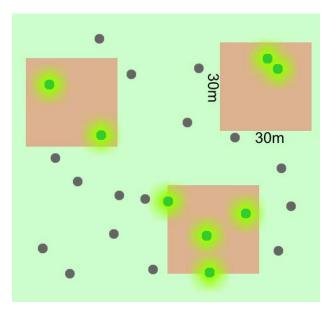


Figure 3. Quadrat count survey method.

Total Count

The fourth method used to inventory burrows is a total or comprehensive count (Figure 4), and it differs from the other methods in that it does not involve sampling theory. Instead, it is a comprehensive count of the entire area of interest, with the assumption that 100 percent of the burrows are observed. Alford (1980), Logan (1981) and McCoy and Mushinsky (1992a) used comprehensive counts for portions of their study areas, while employing strip transects on the larger properties. Though comprehensive surveys are normally considered a technique for smaller areas due to the work involved, comprehensive surveys have been conducted on large areas. A complete listed species census, which included gopher tortoises on $\cong 57,000$ ha of Fort Benning in Georgia, took a team of field workers 3 years to complete (Sandy Abbott, Biologist, U.S. Fish and Wildlife Service, personal communication, 2 March 2004). Comprehensive burrow counts have also been conducted on large private holdings. For example, 7,100 ha of International Paper Company holdings in Mississippi have been completely censused for tortoise burrows (Will McDearman, 23 February 2004).

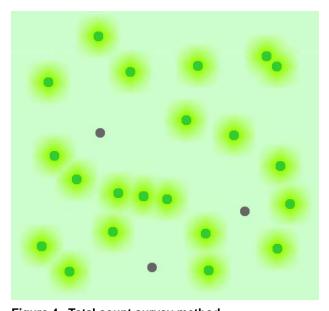


Figure 4. Total count survey method.

Ed Wester (Consulting Biologist, Southern Ecosystems Research, personal communication, 6 February 2004) found that 6 or 7 people walking abreast was an efficient team size for comprehensive searches in large areas. Transect borders were marked with flagging to ensure complete coverage. A South Carolina census was conducted with a team of 60 to 70 volunteers walking abreast (S. Bennett, Herpetologist, South Carolina Department of Natural Resources, personal communication, 6 February 2004). Each observer was instructed to count only those burrows located be-

tween themselves and the person to their left to prevent recording the same burrow twice.

In a Mississippi survey on DeSoto National Forest and on Camp Shelby, observers were spaced 10 to 23 m apart, depending on vegetation density (Mann 1993). Mann (1993) suspected that a previous survey on the study areas had underestimated the number of subadult tortoises because burrows were overlooked because observers were spaced too far apart (20 to 30 m). Mann (1993) also noted that previous surveyors had flagged armadillo burrows as tortoise burrows. The magnitude of misidentification was considered significant for 2 of the 20 sites Mann (1993) studied.

Burrow Activity Classification

A now standard and relatively subjective practice that was started by Auffenberg and Franz (1982) is to guess if a burrow is occupied or vacant based on the external characteristics of the burrow entrance and mound. The guesses are usually on a three- to four-part scale of increasing probability of occupancy (Auffenberg and Franz 1982, Cox et al. 1987, Diemer 1992b, Mushinsky and McCoy 1994, Guyer and Hermann 1997, Tuberville and Dorcas 2001). At the lowest level are burrows with entrances blocked by soil, vegetation, or debris, or burrows that have been taken over by armadillos. These burrows are usually referred to as "old," "closed," "collapsed," "derelict," or "abandoned," and they are assumed empty. The next level on the occupancy scale describes burrows with clear entrances having a well-defined half moon shape, but without fresh tortoise tracks. These burrows are usually referred to as maintained but "inactive," and there is some degree of confidence that the burrow is occupied. If tortoise tracks are present on the mound or in the burrow, the burrow is normally referred to as "active." These burrows are assumed to have the highest probability of occupancy among all burrow classes; however, it is still recognized that the burrow may be vacant for one of three reasons; (1) the tortoise may be above ground foraging or traveling, (2) the tortoise may have moved to another burrow, or (3) the tracks may have been made by a visiting tortoise that lives elsewhere. In addition to these three categories of activity, Diemer (1992b) used a fourth labeled "possibly active" to distinguish burrows with fresh tortoise tracks from those with old tracks. In terms of probability of occupancy, it would fall between inactive and active.

External burrow characteristics can be deceptive indicators of occupancy (Smith et al. 2005). In winter, when tortoises are mostly dormant, fallen leaves can accumulate in burrow entrances, branches can fall on the mounds, and heavy rains can wash soil into the burrow entrances. Any of these can cause an occupied burrow to

appear abandoned. In summer, even a light rain can erase tracks from loose sand, causing a burrow in active use to appear inactive. Tracks from other species traveling in the burrows or walking the mounds can be misread as tortoise sign. In habitat grazed by cattle, trampling can greatly alter burrow appearance leading to inappropriate assessments of burrow status.

Witz et al. (1991) classified 1,019 burrows in central Florida (454 active, 449 inactive, and 116 abandoned) during a summer study when tortoises are active and conditions are appropriate for reading sign. The burrows were excavated after being classified, and occupancy determined. Most of the active burrows were occupied (75 percent), and substantially fewer of the inactive burrows were occupied (12 percent). These results corresponded to expectations. The burrows classified as abandoned were also excavated with unexpected results: 5 percent contained tortoises. The data gathered by Witz et al. (1991) confirm that the external characteristics of the burrow can provide meaningful yet imperfect information on burrow occupancy. There are a few other examples in the literature in which the external characteristics deceived biologists. For instance, Breininger et al. (1991) excavated 10 active burrows, all of which were vacant. A more dramatic example of this was reported by Brandt et al. (1993). They excavated 73 burrows (57 active, 16 inactive), none of which contained tortoises. The authors did not speculate as to why they were fooled, but it is possible that the tracks of burrow commensals were misinterpreted as tortoise sign.

In practice, the most important distinction made in the field is between old burrows and maintained burrows. Maintained burrows include the categories inactive, possibly active, and active, and they are assumed to be potentially occupied. With the exception of Witz et al. (1991), who tested the assumption that old burrows were vacant, most studies accept without challenge that old burrows are empty, and these burrows are subtracted from the total burrow count. The subjectivity of making assessments as to whether or not a burrow is old was measured by Smith and others (2005). Five experienced observers were asked to classify the activity status of 95 tortoise burrows. Depending on observer, the number of abandoned burrows among the 95 was either 10 (10.5%), 25 (26%), 33 (35%), 38 (40%), or 46 (48%). It is obvious that these different interpretations could alter population estimates based on burrow counts, and efforts must be made to more objectively evaluate occupancy status.

Studies Comparing Burrow Counting Methods

Doonan (1986) compared burrow counts using a total count, strip transect, and line transect on a 19.43-ha site in central Florida. Ninety one burrows were found in the

total count (4.68 burrows/ha). The estimated density using 20 randomly located, 150-m x 7-m strip transects was 6.67 burrows/ha (variance was not reported). Line transects (Burnham et al. 1980) were walked down the centerline of each of the 20 strip transects, and the estimated density was 7.76 burrows/ha.

These methods were also compared on a 10-ha study site in northern Florida subdivided into 1-ha sections (Epperson 1997, Doonan and Epperson 2001). The density from the total count was 17.5 burrows/ha (SD 8.54). The density using 10 strip transects (100 m x 20 m, 1 per ha) was 15.5 burrows/ha (SD 10.92) — the strips were searched by 3 observers walking abreast. The difference in densities obtained from total counts and strip transects was not statistically significant. Line transects were walked down the centerline of each strip transect by three observers working independently. Each observer saw a different number of burrows and each count resulted in a different estimate, as follows: 25 burrows observed; 20.9 burrows/ha (18.16 SD); 27 burrows observed; 22.3 burrow/ha (8.23 SD); and 39 burrows observed; 31.4 burrows/ha (21.49 SD). The difference between observers may have been due to sighting aptitude, but it may also have been because one observer inappropriately counted burrows that were noticed from vantage points other than the transect, a violation of survey protocol (Epperson 1997, Doonan and Epperson 2001).

Mann (1993) surveyed 417 ha on 20 sites in Mississippi using a total burrow count and line transects. The results varied by site. On 13 sites, the estimated number of burrows using the line transect method (1,553 burrows) was 49 percent higher than the total count (1,043 burrows). On seven sites, the total burrow count (247 burrows) was 32 percent higher than the line transect estimate (187 burrows). On one site, the counts were equal (12 burrows). The estimated abundances were not statistically compared.

Burke and Cox (1988) compared burrow density estimates using strip transects (250 m x 20 m) and 12 line transects. Three habitat types were surveyed, each with four transects; the line transects were run down the centerline of the strip transects. The authors indicated that the density estimates were comparable for the two more open habitats (5.5 versus 5.3 burrows/ha, and 3.8 versus 3.8 burrows/ha), but that the line transect estimate was more than double the strip transect method for a densely vegetated area (3.3 burrows/ha versus 7.9 burrows/ha). Statistical comparisons were not reported.

The results from the majority of these studies suggest a tendency when using line transects to overestimate abundance in comparison to strip transects and total counts (Doonan 1986, Burke and Cox 1988, Mann 1993, Epperson 1997, Doonan and Epperson 2001). However, burrow detectability was not assessed in any of these

studies, and without knowing the number of burrows that were missed in the strip transect and total counts, it is hard to determine whether line transects overestimate abundance, or if all of the methods underestimate abundance. Burke and Cox (1988) suggested that 20-m wide strip transects underestimated abundance in dense habitat due to missed burrows, thus implying that the line transect method was more accurate under those conditions. Mann (1993) recognized that burrows were probably missed in the total counts, but he believed that the number of missed burrows was insignificant, and that total counts were more accurate than line transect estimates. There is a second problem in interpreting two of the Florida studies because the researchers knew where some of the burrows were located before they conducted the line transect counts (Doonan 1986, Epperson 1997, Doonan and Epperson 2001). This violated survey protocol and it may have altered the results.

Techniques to Convert Burrow Numbers to Gopher Tortoises Numbers

The second step in the estimation process is to convert the burrow count to a tortoise count. The methods have been reviewed by Cox et al. (1987), Burke and Cox (1988), Burke (1990), Epperson and Heise (2001), and Moler and Berish (2001).

The original method to convert burrows to tortoises was developed by Auffenberg and Franz (1982), and it required no field work other than a burrow survey. The method was based on a study of 122 burrows over a ≤15-year period, during which, on average, 61.4 percent of the maintained burrows were occupied (Auffenberg and Franz 1982). The method assumed that all old burrows were empty. The categories of active and inactive were combined into one, which was multiplied by 0.614 to convert from burrow abundance to tortoise abundance. Auffenberg and Franz (1982) applied this rate to convert burrow numbers to tortoise numbers for data collected across the southeastern United States. They referred to the occupancy rate as a correction factor, a term since adopted in the gopher tortoise literature.

The Auffenberg and Franz (1982) correction factor tends to overestimate abundance (McCoy and Mushinsky 1992b, Moler and Berish 2001), and several biologists have commented on the risks of using this or any other rate without on-site verification since the tortoise/burrow ratio may change by site, season, and year (Burke and Cox 1988, Burke 1989, Breininger et al. 1991, McCoy and Mushinsky 1992b, Moler and Berish 2001).

McCoy and Mushinsky (1992b) worked to improve on the approach of Auffenberg and Franz (1982) by modeling burrow and tortoise counts from 26 areas. Separate regression formulas were calculated to convert active burrows to tortoises and to

convert active plus inactive burrows to tortoises. Their methods were considered a substantial improvement over the use of a single conversion factor, but the formulas were not considered accurate enough by Moler and Berish (2001) for valid estimates under all conditions.

Moler and Berish (2001) considered using burrow counts as an index of abundance, thus skipping the conversion altogether, but since the ratio of burrows/tortoise can vary due to factors other than tortoise abundance (such as habitat quality and social interactions that affect tortoise movements), they concluded that burrow counts without supporting data on occupancy could give false impressions regarding population trends. For instance, as habitat quality declines, tortoises increase their movements, and increase the number of burrows they use in exploiting larger home ranges. If a survey documented an upward trend in burrow use, it would likely be interpreted as evidence that there were more tortoises, when in fact the numbers might be stable. Social factors can also influence burrow use, thus altering the tortoise/burrow ratio. It is conceivable that a population reduction of adult female tortoises due to illegal harvest could lead to increased movements by males searching for females. Burrow use could actually increase in this situation, giving the impression of higher tortoise numbers, when the reality would be that there were fewer tortoises in the population. Reasons such as these are why Moler and Berish (2001) cautioned against using burrow surveys as indices of abundance.

In surveys that have determined on-site occupancy rates, the most common technique used is the burrow camera (Spillers and Speake 1988, Mann 1993, Tuberville and Dorcas 2001, Hermann et al. 2002). The cameras are valuable tools, but they are not perfect. Breininger et al. (1991) tried to scope flooded burrows, but the water was too turbid to see. Hermann et al. (2002) reported camera results with the qualifier that occupied burrows contained at least one tortoise. This was in recognition that burrows may contain two tortoises in single file, and because of the tight fit, there is insufficient room to maneuver the camera past the first tortoise. Double occupancy is not often seen: it was not observed by Witz et al. (1991) when they excavated 1,019 burrows containing 400 tortoises, and it was observed by Smith et al. (1997) for only 10 of 1,344 radio locations of 14 tortoises. A separate problem with camera surveys is that not all burrows can be scoped. The camera used by Burke (1989) could fit only in adult-sized burrows. In Mississippi, Mann (1993) successfully scoped most (83.6 percent) of the 353 burrows that were attempted; occupancy could not be determined for 58 burrows due to obstructions or convoluted tunnels; in 23 burrows of these burrows, a wall of dirt near the bottom of the burrow stopped the camera, and since the wall may have hidden a tortoise, occupancy could not be ruled out. Kent et al. (1997) used a camera to determine occupancy in 202 of 208 burrows attempted; the unsuccessful attempts were because the burrows were too long for the equipment (n=6), the burrow was too twisted to navigate (n=1), and the

burrow collapsed during inspection (n=1). The camera head was only 0.9 cm diameter, and the authors indicated the camera was used successfully in small burrows, which is likely given that hatchling burrows average 5 cm wide by 3 cm tall (Doonan and Stout 1994). Apparently no problems were experienced with the camera pushing dirt over a small tortoise and obscuring them from sight, which was a potential problem mentioned by Breininger et al. (1991).

A second method used to estimate occupancy rate is to capture the tortoises in pit-fall or cage traps set at the burrow entrance. Trapping requires an extensive commitment since tortoises can take weeks to emerge from their burrows, and the traps must be checked daily. Trapping using pitfall traps can be destructive to nests located in the burrow mound, a problem that can be avoided using cage traps. Cage traps have the drawback of greater expense and bulk over the plastic buckets used for pitfall traps, and they may be less efficient at catching tortoises. Numerous projects have estimated tortoise abundance from trapping (Linley 1986, Doonan 1986, McCoy and Mushinsky 1992b, Diemer 1992b, Smith 1992). Trapping is the best method for projects requiring information that can be gathered only by handling tortoises, such as sex ratios and health assessments.

The third, less direct method to determine burrow occupancy is to monitor tortoise traffic using a miniature fence of sticks placed in the burrow mouth, and watching over time to see if a tortoise knocks down the fence, thus indicating use. This old-fashioned method was originally used by Hallinan (1923) to monitor daily activity patterns of tortoises. It requires repeated visits to the burrows to monitor traffic, and observers skilled at reading sign. Burrow commensals may confound efforts by disturbing the sticks. Forked sticks have been used to distinguish between tortoises leaving the burrow from those moving in (Breininger et al. 1991).

Burke and Cox (1988) describe an assortment of other methods used to determine whether a burrow is occupied. These include observing the direction of tortoise tracks in the burrow, listening for breathing or movement from the burrow, and the insertion of flexible wires and pipes to feel the tortoise. Some tortoises can be lured out of their burrows by slapping the burrow mound with the palm of your hand. These methods all work on occasion, but none reliably enough for abundance estimates.

Studies Comparing Techniques to Determine Burrow Occupancy

Burke (1989) compared burrow occupancy using a camera and the stick method during a June survey in southern Georgia. Sticks were placed in the entrances of 199 burrows to record burrow traffic; tortoises used 73 (36.7 percent) of the burrows.

The camera was used on 22 randomly selected burrows; 6 (27.3 percent) were occupied. The reason for the higher estimate using the stick method was not noted. Burke (1989) concluded the stick method as employed in the study was appropriate for coarse estimates of occupancy, but the camera was more accurate.

Breininger et al. (1991) used bucket traps, sticks, burrow excavation, a burrow camera, and an experienced gopher tortoise hunter to estimate burrow occupancy rates in randomly located 50-m x 30-m plots. The hunter used a pulling hook and the direction of fresh tortoise tracks to assess occupancy. The occupancy rate for active burrows varied from 0 percent to 79 percent depending on method and time of year. The rate for inactive burrows varied from 0 percent to 7 percent. Breininger et al. (1991) concluded that burrow cameras were the best method in terms of cost and accuracy.

Burke and Cox (1988) tested if the direction of tortoise tracks in the burrow was reliable in assessing burrow occupancy. An experienced tortoise hunter, after studying tracks at burrow entrances, judged that 34 burrows were occupied. The actual number of burrows occupied was 15, a 41 percent accuracy rate.

Smith and others (2005) tested the accuracy of burrow cameras in determining burrow occupancy on Merritt Island, Florida. Two teams, each with 2 observers, scoped 57 burrows. Each burrow was subsequently excavated and the camera results verified. The excavations revealed that 11 burrows contained 1 tortoise each. One team saw all 11 tortoises using the camera, and the second team saw 8 of the tortoises. The burrows were reportedly straight tunnels, and relatively easy to scope with a camera. The reasons why the second team missed three tortoises were not reported.

Potential Accuracy of Techniques

The accuracy of the various methods for estimating burrow abundance and burrow occupancy have been discussed by the following authors: Burke and Cox (1988), Breininger et al. (1991), Diemer (1992b), Mann (1993), Kent et al. (1997), and Moler and Berish (2001). Several general conclusions can be reached. One is that none of the tests comparing methods have been extensive enough in scale or detail to be considered conclusive. A second is that it appears that any of the techniques have the potential to accurately determine abundance of adult tortoises if vegetation is sparse, the surveyors experienced and motivated, the sampling unbiased, the analysis appropriate, and the burrow occupancy rates correctly measured. If conditions are otherwise — poor visibility, unmotivated observers, biased sampling, inadequate sample size, inappropriate analysis, and incorrect occupancy rates — any of

the techniques have the potential to be well off the mark in an unpredictable direction. The major sources of error that have been identified include overlooked burrows, misidentified burrows, misclassified burrows, overlooked tortoises, incorrect correction factors, and biased sampling. Although most surveys make an effort to minimize error, to date there has been no standardized attempt to quantify the error for use in calibrating survey results.

Detectability

Virtually all methods of population estimation are based on count statistics (e.g., number of animals or burrows detected, number of animals captured). Such counts generally represent an unknown fraction of the total population of animals (or burrows), and additional information is usually needed for estimating abundance or making inferences regarding the estimates of abundance. In particular, the fraction of the total population represented by count statistics (or equivalently, fraction of total population not detected) must be estimated (Williams et al. 2002). Detailed discussion of detectability (probability of detecting an animal or a burrow if it is present in the study area) and other issues involved in abundance estimation are in Williams et al. (2002); a brief overview follows.

Suppose that a habitat patch is surveyed for gopher tortoise burrows, and that the only burrows not included in the count are those not detected by the observers (i.e., all of the study area was surveyed). Let C be the resulting count statistic (i.e., number of burrows detected), and β be the probability of detecting a burrow, given that it is present in the study area. The expected value (E) of the count statistic is:

$$E(C) = \beta N$$

where N is the actual population size. We can think of β as the fraction of the burrows that are present but not detected. If the exact value of β is known, this information can be used to obtain an unbiased estimate of population size N as:

$$\hat{N} = \frac{C}{\beta}.$$

In practice, however, β is rarely known, and an important goal of most abundance estimators is to estimate this quantity. Using the estimate β , an unbiased estimate of N can be obtained:

$$\hat{N} = \frac{C}{\hat{\beta}}.$$

As an example, suppose that we count 100 tortoise burrows (C = 100) during a survey of a gopher tortoise habitat, and that we assume the detection probability to be 0.25 ($\hat{\beta}$ = 0.25). Then, an unbiased estimate of the number of burrows in the study site is 100/0.25=400. Note that C = N if and only if $\hat{\beta}$ = 1.0, which is rarely the case. Finally, note that virtually all abundance estimators attempt to estimate θ in some ways. Estimates of population size based on the assumptions of θ = 1 may not be trusted without convincing evidence that every animal or burrow present in the study area was detected.

This formal challenge of the assumption that all burrows are observed, and efforts to measure burrow detectability for each burrow size class (juvenile, subadult, and adult [i.e., small, medium, and large]), traditionally have been lacking from burrow surveys and are needed to increase accuracy in total counts and in any of the sampling methods using plots (J. Nichols, Senior Scientist, Patuxent Wildlife Research Center – USGS, personal communication, 8 March 2004). Detectability is partially a function of burrow/mound size, and a separate estimate of detectability is appropriate for each size class. Habitat quality and density of vegetation are also important considerations. One approach to estimating detectability is the double observer method (J. Nichols, 8 March 2004; Williams et al., 2002), wherein an area that has been surveyed is resurveyed by a second observer(s), and the number of burrows missed in the first effort is used to calibrate the results from the first survey. The resurvey need cover only a random sample of the first area. Estimates of burrow detectability by size class, in addition to improving burrow surveys for abundance estimates, would also add confidence to estimates of reproduction and recruitment. These assessments are a common element of burrow surveys, but there is always the concern that they do not accurately reflect the true population structure since it is likely that burrows are missed in proportion to size. This skews the observed structure toward the larger size classes, and an underestimation of recruitment.

Stratification

Areas of public and military lands where estimations of gopher tortoise abundance may be needed are usually sufficiently large enough to have considerable heterogeneity in habitat types and land use. An important component in the design of efficient and accurate surveys for these locales is stratification — the use of existing knowledge of the study area to subdivide the area into sections (strata) with similar attributes (habitat qualities, tortoise density, etc.). Estimates of total population

size are then obtained by combining stratum-specific estimates (Williams et al. 2002). This helps to reduce sample variance (increases precision). And subdividing the study area increases the likelihood that random samples (transects, quadrats) will give an accurate picture of the situation. Without stratification, it would be possible to randomly select samples that are clumped in a portion of the study area. If that portion of the study area is not representative (high density, low density, etc. in comparison to the total area), the sample data would give a misleading picture of the situation.

Cochran (1977) suggests construction of strata based on the frequency distribution of the variable in question or some highly correlated variable, therefore stratification is most easily addressed on sites where prior surveys can provide information on spatial occurrence of tortoise aggregations and/or suitable habitat types. A stratification plan should be designed by personnel with a good knowledge of sampling, working closely with persons highly familiar with the area (resource managers). This aspect of survey preparation has not been particularly well elucidated in the existing gopher tortoise census literature, yet better estimates can be obtained by focusing the sampling effort.

Simulation Modeling With Existing Data

As previously mentioned, sites with available information on distribution of species of interest and habitat types lend themselves more readily to stratification. In the course of this review of tortoise survey techniques, data was obtained from two military installations where total tortoise counts had been carried out:

- Fort Benning, Georgia GIS of tortoise burrows, vegetative cover and soil types
- Camp Shelby, Mississippi GIS of tortoise burrows and soil types

Review of these data sets indicated that they provide habitat and distribution information suitable for delineating future sampling strategies, and also suggested their suitability for investigative modeling of sampling techniques.

A simple line transect simulation model was created with MATLAB^{TM*} using the tortoise burrow data from Fort Benning, to examine the effects of varying number,

* Citing trade names does not constitute endorsement by the Department of Defense or the U.S. Army. MATLAB is a product of The MathWorks, 3 Apple Hill Drive, Natick, MA 01760-2098; http://www.mathworks.com/

length and orientation of transects. To calibrate the detectability term in the model, some field tests measuring the distance of observable burrows from transect lines were carried out at the Katherine Ordway Nature Preserve in Alachua County, Florida. The modeling approach has proven interesting, providing abundance estimates approaching actual count numbers. Continuing work includes incorporation of the soils and vegetative cover data into the decision process on allocation of transects. Strip transect, quadrat and adaptive cluster sampling allocation may benefit from similar evaluation, as might comparison of the techniques. This general methodology is being widely evaluated in the field of abundance estimation, and in this case promises utility upon further reparameterization and calibration of the model with site-specific information.

Summary and Implementation of Abundance Estimation Techniques

Total Counts

A total burrow census may be the best choice in situations when the goal is identify all tortoise burrows in the area or to confirm the absence of burrows. However, it should be noted that burrows can be overlooked using a total count approach, and as with the sampling techniques discussed below, all tortoise survey results will be improved upon if detectability estimates became a routine part of all enumeration techniques. The U.S. Fish and Wildlife Service requires total counts in the federally listed portion of the tortoise's range to assess habitat alterations that might negatively affect the species (Will McDearman, 23 February 2004). Total counts have the advantage of not requiring time in the set-up and design of survey protocol since the entire area will be investigated. Labor in terms of time and manpower is the main drawback of total counts, and is generally viewed as unsupportable for large-scale population monitoring. As a labor-saving measure, Moler and Berish (2001) suggest using permanent "sentinel sites" of 20 to 40 ha, within which total counts are conducted periodically.

Sampling Techniques

The alternative to total counts is a sampling approach. In situations when sampling will answer the questions, quadrat sampling is probably the most versatile method to census tortoise burrows. It can be used in any habitat type, the statistics are straightforward, and if the plots are randomly located, the estimates unbiased. Long, thin plots are probably a better choice in most situations than square plots. Quadrats with flagged corners and boundaries lend themselves easily to measurements of detectability.

The strip transect method is also versatile; however, it is inefficient in dense habitat due to narrow transect width. In open habitat, with use of double observers and accurate measurements of transect length and width, it would be an efficient sampling method.

The line transect method has an advantage over the strip transect method in that burrow detectability is not assumed to be 100 percent. The lack of this assumption results in more area covered per transect length per observer, and it eliminates the need to measure burrow detectability. The disadvantages of the line transect method are due to the added time involved in taking measurements from transect to burrows, to the sample size requirements of >40 burrows (which limits its use to larger areas), and to the complexity of the DISTANCE software program. The estimates produced by DISTANCE depend on choices made by the user, and if the wrong choices are made due to lack of expertise with the program and the method, incorrect answers will result. Given the potential problems associated with DISTANCE, its use is not recommend until further testing demonstrates its value.

None of the methods used to date may work well in terms of cost or statistical efficiency in habitat with low density, clumped populations. Under these conditions, a method called adaptive, or cluster sampling with randomly located plots may have merit (Thompson 1992). The idea behind the method is that searches should be more extensive in areas surrounding occurrences, and less intensive elsewhere. Once an occurrence is documented in a plot, the neighboring plots are searched. No additional searches are made next to empty plots. The method yields unbiased estimates of abundance, with reportedly greater efficiency than other sampling methods under conditions of rareness and clumping (Thompson 1992). The general search strategy in adaptive sampling has been used in Mississippi since the mid-1990's to delineate the boundaries of tortoise colonies by searching outward 284 m from active burrows (W. McDearman, 25 March 2004). The searches continue outward in 284-m belts until no additional burrows are found. The method was first used in conjunction with strip transect sampling, with expanding searches used around plots with active burrows. It is currently used to delineate colony boundaries in development sites.

Occupancy Rates

Keep in mind that there are many situations when estimates of occupancy are not needed, and in these cases, there is no reason to convert accurate burrow counts to tortoise estimates of questionable accuracy. For instance, burrow surveys without conversion to tortoise estimates are adequate to determine presence/absence. Burrow surveys with GPS locations can be used to map distributions, and changes in distributions can be used for coarse assessments of population status. Coarse as-

sessments of population change can also be made using trends in the number of maintained burrows, although there are many pitfalls to this approach (Moler and Berish 2001). Burrow surveys also have the potential to measure reproduction and recruitment if accurate burrow measurements are taken during the surveys, and the searches are intensive for smaller burrows. The earlier suggestions to assess burrow detectability by size class will greatly aid this aspect of burrow surveys.

In situations when accurate tortoise abundance estimates are needed, more rigorous estimates should be employed. Burrow cameras appear to be the most practical tool to measure occupancy. Burrow camera surveys have an advantage over other methods in that they can be used 12 months of the year, thus permitting winter surveying, whereas the other methods are only practical when tortoises are active (about 5 to 7 months of the year).

Trapping and sticks are two other options to determine burrow occupancy. The stick method is simple, and the use of forked sticks (or some other device that indicates direction of travel) may be more accurate than previously shown if the observer is skilled at reading sign and the burrows are checked daily. The number of observation days has not been determined, but this would not be a difficult research problem. Trapping is the most labor intensive method to verify occupancy. Unless data are needed that requires handling tortoises (sex, reproductive status, health assessment, etc.), there are better ways to measure occupancy.

There were no examples in the literature where trail cameras were used to measure occupancy rates, and these cameras may have advantages over other methods. They can be quickly deployed (less than 5 minutes), and the photographs require no special skill to interpret, unlike field sign. Tortoises will not be overlooked as is possible with burrow cameras. Trail cameras require periodic monitoring to change film, but if activity is light, a roll of film with 36 exposures would last longer than 1 week. One disadvantage is that a large number of cameras would be required (>20) to cover an entire colony which often contains more than 72 individuals, and loss/tampering may be a problem in some areas. Guyer et al. (1997) built pressure pad-actuated cameras that were subsequently used for studies of tortoise behavior (Boglioli et al. 2000); light-beam interruption and intervalometer-actuated cameras are available commercially.

Regardless of the method chosen to determine occupancy, occupancy rates should be determined for each burrow category, rather than making assumptions with unknown validity. At least two categories seem appropriate for surveys: maintained burrows and old burrows. The term "abandoned burrow" should probably be avoided since it can be a misnomer (Witz et al. 1991). Since classifications are based on evidence of tortoise activity, observers should pay attention to season as it

relates to tortoise activity, precipitation as it influences tortoise sign, and human or animal activity that might alter burrow appearance. There will always be some level of judgment involved in assigning burrows to categories, and some burrows will fall in the gray area between categories. Observers must be able to distinguish tortoise burrows from armadillo burrows. If there is any question, Mann (1993) suggests that all burrows be measured (tortoise and armadillo), and that burrows with height and width equal be discarded from the results (armadillo burrows are round, whereas tortoise burrows are half circles that are about twice as wide as they are high).

3 Conclusions and Next Steps

The aforementioned versatility of the quadrat (plot) sampling method recommends it as a default technique that can be used in many sampling circumstances with reasonable expectations of accuracy when carried out properly. Strip transects with reasonable estimates of detectability can offer similar utility when habitat conditions make them appropriate. The foregoing literature review clearly illustrates the voids in our knowledge about the accuracy of current gopher tortoise abundance estimation techniques. Conceivable worst-case scenarios could be acceptance of surveys with unchallenged assumptions of total observability of burrows, followed by conversion of those survey results to tortoise numbers utilizing published correction factors that may not be applicable to the survey site.

As discussed earlier, previous studies comparing burrow survey methodologies have not adequately addressed the issue of detectability, which presents a weakness in current population estimation and total count techniques. It is therefore recommended to undertake a rigorous field test of these techniques to remove the detectability weakness. This validation would involve first applying the sampling methodologies (quadrats, strip transects, and line transects) on an appropriately sized area where burrows have been undisturbed and not visibly marked, utilizing an experienced team of technicians. The sampling would be followed by total counts; this will allow rigorous estimates of detectability. The critical final step would be estimation of burrow occupancy rates using a burrow camera. Using an "undisturbed" area and progressing from the sampling techniques through the total counts to burrow occupancy will:

- eliminate many of the potential sources of bias that would arise if total counts were performed first,
- provide method-specific estimates of detectability, including evaluation of the hitherto uncontested assumption of 100 percent detectability in total counts,
- allow assessment of the relative accuracy of the sampling methods when carried out in a similar time and area,
- allow evaluation and comparison of the efficiency of application between the different methods when carried out in a similar time and area, and
- provide site-specific information on occupancy to assist in deriving estimated tortoise abundance from burrow counts.

This systematic field validation of abundance estimation techniques will be highly useful in providing natural resource managers with defensible information and cri-

teria upon which to base decisions on efficient modes of action. Additionally, systematic field validation of abundance estimation techniques would lead to realization of increased accuracy for surveys on installations, while also making a valuable contribution to gopher tortoise population abundance estimation.

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14. ABSTRACT

Estimating threatened and endangered species (TES) population trends is essential to be able to track a species' recovery progress. Natural resources managers on military installations use a wide variety of survey and monitoring methods, with incomplete knowledge toward their accuracy. Using the gopher tortoise (*Gopherus polyphemus*) as the test case species, this report examines TES population estimation techniques for accuracy. Managers are cautioned to not accept surveys with unchallenged assumptions of total observability of burrows, and to not convert survey results to tortoise numbers utilizing published correction factors that may not be applicable to a local survey site.

The versatility of the quadrat (plot) sampling method recommends it as a default technique that can be used in many sampling circumstances with reasonable expectations of accuracy. Strip transects with reasonable estimates of detect-ability can offer similar utility when habitat conditions make them appropriate.

Previous studies comparing burrow survey methodologies have not adequately addressed the issue of detectability, which presents a weakness in current population estimation and total count techniques. It is therefore recommended to under-take a rigorous field test of these techniques to remove the detectability weakness.

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